



UNIVERSITI PUTRA MALAYSIA

**GENETIC STUDIES OF BACTERIAL WILT RESISTANCE AND
AGRONOMIC CHARACTERS, AND YIELD COMPONENT
ANALYSIS IN TOMATO**

SAAD OSMAN ABDALLA

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By

SAAD OSMAN ABDALLA

**Thesis Submitted in Fulfillment of the Requirements for the Degree of
Master of Agricultural Science in the Faculty of Agriculture, Universiti
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DEDICATION

To,
my father, mother, and three sisters



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TABLE OF CONTENTS

	Page
ACKNOWLEDGMENTS.....	iii
LIST OF TABLES.....	viii
LIST OF FIGURES.....	xi
ABSTRACT.....	xii
ABSTRAK.....	xiv
CHAPTER	
I INTRODUCTION.....	1
II LITERATURE REVIEW.....	5
Bacterial Wilt of Tomato.....	5
Breeding for Bacterial Wilt Resistance.....	7
Genetic Control of Bacterial Wilt Resistance.....	9
Mechanism and Factors Affecting Bacterial Wilt Resistance.....	10
Breeding Tomato for Disease Resistance.....	13
Breeding Tomato for Heat Tolerance.....	15
Breeding for Yield and Yield Components.....	18
Heterosis.....	19
Heritability.....	26
Correlation between Characters.....	29
Path Coefficient Analysis.....	31
Regression Analysis.....	32
Tomato Research in Malaysia.....	33



III	MATERIALS AND METHODS.....	35
	Experiment 1: Greenhouse Screening Trial.....	36
	Preparation of Planting Materials.....	36
	Inoculation Procedures.....	37
	Experiment 2: Field Screening Trial	38
	Hybridization for F ₁ Seeds Production.....	39
	Production of F ₂ and Backcrosses Seeds.....	41
	Experiment 3: Second Greenhouse Screening Experiment.....	42
	Experiment 4: Hydroponic Evaluation.....	45
	Measurement of Characters.....	49
	Statistical Analysis.....	51
	Heterosis Estimation.....	51
	Analysis of Variance Procedure.....	51
	Heritability Estimates.....	53
	Correlation Analysis.....	54
	Path Coefficient Analysis.....	55
	Regression Analysis.....	56
IV	RESULTS AND DISCUSSION.....	58
	Bacterial Wilt Resistance.....	58
	Performance of Parents, F ₁ s, F ₂ s, and Backcrosses for the 12 Characters.....	66
	Heterosis.....	91
	Heritability.....	100
	Correlation between Characters.....	105
	Path Coefficient Analysis.....	110
	Regression Analysis.....	117
V	SUMMARY AND CONCLUSIONS.....	125
	BIBLIOGRAPHY.....	128



APPENDICES	144
A Additional Tables	145
B Bar Charts	147
C Computer Programmes	161
VITA	178



LIST OF TABLES

Table		Page
1	Names, designations, growth habits, and some remarks on the six tomato varieties used in the study	35
2	The selected cross combinations (populations) of tomato varieties used in the experiments	43
3	Selected populations of the tomato varieties with their corresponding number of plants for each genotype used in the second screening experiment (Experiment 3)	44
4	Nutrient elements used to prepare the nutrient solution in the hydroponic evaluation experiment (Experiment 4)	58
5	ANOVA table based on plot means (RCBD).....	52
6	ANOVA table based on individual observations (CRD).....	53
7	Means and variances of the disease indexes of the tomato six varieties grown in the greenhouse (Experiment 1).....	59
8	Percentage survival of plants with their respective values angular transformation for the six tomato varieties in the field screening (Experiment 2)	60
9	Means and variances of disease indexes of the three tomato populations observed in the second greenhouse screening experiment (Experiment 3)	61
10	Susceptibility of bacterial wilt. Ranking of the 17 cross combinations of tomato according to their disease indexes ...	63
11	Percentage increase or decrease of F_1 s over mid-parent and better-parent values and inbreeding depression of the disease indexes of the three tomato populations observed in the greenhouse (Experiment 3)	64
12	Broad and narrow sense heritability estimates of the disease indexes in the three tomato populations observed in the greenhouse (Experiment 3)	65



13	Mean squares and error mean squares from the analysis of variance for the 12 characters of tomato varieties studied in the hydroponics (Experiment 4)	67
14	Mean values for the 12 characters measured on the five tomato populations in Experiment 4	69
15	Variance estimates of the 12 characters measured on the five tomato populations in Experiment 4	80
16	Coefficient of variation percentages (CV%) of the 12 characters measured on the five tomato populations in Experiment 4.....	86
17	Percentage increase or decrease of the F_1 values over mid-parent (upper row) and better-parent (middle row) and inbreeding depression percentages (lower row) of the 12 characters measured in the five tomato populations studied in Experiment 4.....	92
18	Broad sense (upper row) and narrow sense (lower row) heritability estimates of the 12 characters measured on the five tomato populations.....	101
19	Simple phenotypic correlation coefficients between the 12 characters measured on tomato	106
20	Path coefficient analysis showing the direct and indirect effects (P value) of some characters of tomato on yield.....	111
21	Linear regression analysis between yield and other characters of tomato	118
22	Summary of forward selection procedure for the dependent variable yield of tomato	120
23	Summary of backward elimination procedure for dependent variable yield of tomato	120
24	Analysis of variance table for regression analysis with stepwise selection procedure at 0.15 probability level (Yield is the dependent variable)	122
25	Summary of stepwise procedure for dependent variable yield of tomato	122



26	R ² of single and/or combination of different independent variable (s) with dependent variable yield of tomato.....	123
27	ANOVA table of disease index for the first greenhouse screening experiment (Experiment 1)	146
28	ANOVA table of disease index for the second greenhouse screening experiment (Experiment 3).....	146



LIST OF FIGURES

Figure		Page
1	Bar chart showing the disease indexes of the three populations studied in Experiment 3	148
2	Bar chart showing mean values of yield per plant of the five populations studied in Experiment 4	149
3	Bar chart showing mean values of number of fruits per plant of the five populations studied in Experiment 4	150
4	Bar chart showing mean fruit weights of the five populations studied in Experiment 4	151
5	Bar chart showing fruit diameters of the five populations studied in Experiment 4.....	152
6	Bar chart showing days to flowerings of the five populations studied in Experiment 4.....	153
7	Bar chart showing days to first harvests of the five populations studied in Experiment 4.....	154
8	Bar chart showing days to last harvests of the five populations studied in Experiment 4.....	155
9	Bar chart showing plant heights of the five populations studied in Experiment 4.....	156
10	Bar chart showing number of branches per plant of the five populations studied in Experiment 4	157
11	Bar chart showing stem diameters of the five populations studied in Experiment 4	158
12	Bar chart showing fruit pH readings of the five populations studied in Experiment 4	159
13	Bar chart showing fruit total soluble solids (TSS) percentages of the five populations studied in Experiment 4.....	160



Abstract of the thesis presented to the Senate of Universiti Pertanian Malaysia in fulfillment of the requirements for the Degree of Master of Agricultural Science.

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By

SAAD OSMAN ABDALLA

July, 1996

Chairman: Prof. Yap Thoo Chai, Ph. D
(December 1991 - December 1995)

Dr. Hiriyati Abdullah
(December 1995 - July 1996)

Faculty : Agriculture

Experiments were conducted to evaluate the inheritance and performance of six tomato parental lines (varieties) and their respective cross combinations. Inheritance of bacterial wilt resistance was evaluated in the greenhouse (screening experiment), while assessments of twelve agronomic characters were made under hydroponic conditions.

Greenhouse inoculation studies indicated that, parents have different degree of susceptibility to bacterial wilt. There were significant differences ($P = 0.01$) among genotypes as indicated by their disease index. Studies carried out under hydroponic conditions showed that except for the



number of branches per plant and stem diameter, there were significant differences ($P = 0.01$) among the means of all the characters studied.

Different degree of heterosis have been expressed for different characters. For the three populations studied, resistance to bacterial wilt showed high heterosis over mid parental value and no heterosis over better parental value. On the other hand, for most of the agronomic characters in the five populations studied, a majority showed heterosis over better parent, but their values were not high. However, mid parental heterosis values were mostly high.

Heritability estimates were generally high for most characters including disease index. This indicated that the additive genes were more important than non-additive genes.

Yield characters (viz. number of fruits per plant, average weight per fruit and fruit diameter) were highly and positively correlated with each other. Maturity characters (days to flowering and days to first harvest) were in general negatively correlated with yield.

The path coefficient analysis revealed that the number of fruits per plant, average weight per fruit, and fruit diameter were the most important characters that contributed directly to the yield.

Therefore, in any breeding programme selection for disease resistant plants (genotypes) after each generation of selfing should also take into consideration other desirable agronomic characters.



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**KAJIAN GENETIK RINTANG LAYU BAKTERIA DAN SIFAT AGRONOMI, DAN
ANALISIS KOMPONEN HASIL TANAMAN TOMATO**

oleh

SAAD OSMAN ABDALLA

Julai, 1996

Pengerusi: Prof. Yap Thoo Chai, Ph. D
(Disember 1991 - Disember 1995)

Dr. Hiriyati Abdullah
(Disember 1995 - Julai 1996)

Fakulti: Pertanian

Ekspirimen telah dilakukan untuk menilai warisan dan prestasi enam baris induk tomato (varieti) dan kombinasi silang masing-masing. Warisan keresistanan penyakit layu bakteria telah dinilai di rumah hijau (eksperimen penyaringan) sementara penilaian dua belas ciri agronomi telah dikaji di dalam kaedah hidroponik.

Kajian inokulasi di rumah hijau menunjukkan induk mempunyai darjah keresistanan yang berbeza kepada penyakit layu bakteria. Terdapat perbezaan ketara antara genotip seperti yang ditunjukkan di dalam indeks penyakit. Kajian di bawah keadaan hidroponik menunjukkan bahawa kecuali bagi pertumbuhan



dahan setiap pokok dan diameter batang, terdapat perbezaan bererti ($P = 0.01$) di antara purata semua ciri yang dikaji

Heterosis dengan darjah yang berbeza telah ditunjukkan bagi beberapa penyakit layu bakteria yang berlainan. Bagi tiga populasi yang dikaji, ketahanan terhadap penyakit layu bakteria menunjukkan heterosis yang tinggi pada nilai pertengahan induk dan tiada heterosis pada nilai induk terbaik. Selain dari itu, bagi kebanyakan ciri-ciri agronomi yang terdapat di dalam lima populasi yang dikaji, kebanyakannya menunjukkan heterosis keatas induk terbaik tetapi nilainya tidak tinggi. Walau bagaimanapun, nilai heterosis pertengahan induk kebanyakannya adalah tinggi.

Anggaran keterwarisan pada umumnya adalah tinggi untuk kebanyakan ciri-ciri termasuklah indeks penyakit. Ini menunjukkan pentingnya gen tambahan daripada gen bukan tambahan.

Ciri-ciri hasil (bilangan buah setiap pokok, berat purata buah dan diameter) adalah tinggi dan berkolerasi secara positif antara satu sama lain. Ciri-ciri kematangan (hari untuk berbunga dan hari untuk tuaian pertama) pada umumnya berkolerasi secara negatif dengan hasil.

Analisis angkali haluan, membuktikan bahawa bilangan buah setiap pokok, purata berat buah, dan diameter buah adalah ciri yang terpenting yang menyumbang secara langsung kepada hasil.

Dari itu di dalam program baik baka, pemilihan untuk tanaman (genotip) resisten kepada penyakit, selepas setiap generasi haruslah mengambil kira sifat-sifat agronomi yang baik. Pemilihan untuk genotip selepas setiap generasi harus menggabungkan resisten dengan ciri-ciri lain yang dikehendaki di dalam beberapa kombinasi kacukan (populasi).

CHAPTER I

INTRODUCTION

Tomato is one of the most important vegetable crops grown in the world (Gould, 1983).

The cultivated tomato *Lycopersicon esculentum* Mill, belongs to the family *Solanaceae* together with some economic plants like potato, egg plant, tobacco and pepper, which are characterized with the production of an alkaloid substance known as the solanine (Morrison, 1938).

Tomato is indigenous to tropical America, Mexico and the Andes in Peru. The cherry tomato is considered as the original or the wild ancestor of the present cultivated tomato (Morrison, 1938). Tomato was considered an ornamental plant in the early days, where the colour of the fruit was yellow rather than red. Now it is generally accepted as a vegetable crop and has become an important food crop in the 19th century (Gould, 1983 ; Tigchelaar, 1986).

Tomato can be cultivated in different types of climatic conditions indicating the wide genetic diversity of the crop. However, countries with long dry growing season are considered to be the major areas for production of processed tomatoes (Tigchelaar, 1986).



In the United States, tomato occupies 90% of home gardening. In Europe and USA greenhouse cropping plays a major role in the production of fresh tomatoes. In Europe, the yield of tomato is able to reach as high as 135-180 t/ha in glasshouse cultivation (Yamaguchi, 1983)

According to the Food and Agriculture Organization (FAO), in 1991 Europe produced 16,708,000 t of tomatoes, North America 1,202,800 t, the Far East 9,590,000 t and Africa 346,300 t. In Malaysia the production was 7,000 t of tomatoes with an average yield of 17.5 t/ha (FAO, 1992).

Bacterial wilt caused by *Burkholderia solanacearum* (Synonym *Pseudomonas solanacearum*) is an important disease in tomato. It was first recognized by Rolfs (1898), and Earle (1900). The soil-borne bacterial pathogen infects the vascular tissues, producing symptoms of wilting or stunting and yellowing of the foliage, leading to the death of susceptible plants. It is a major disease in areas where the pathogen is well established in the soil (Singh, 1987). In the tropics, the disease had since been reported to cause losses from 30 to 100 % of the crop (Villareal, 1980). In Malaysia, the disease is considered as the most important limiting factor for tomato production (Graham et al., 1977).

B. solanacearum has a wide range of hosts and the common hosts include all the plants in the *Solanaceae* family, bananas, and peanuts. About 33 plant families have been reported to be the host of the pathogen (Kelman, 1953). Several crops and weeds were suspected to be symptomless carriers for the pathogen (Opio, 1983).

Development of tomato varieties or lines which are genetically resistant to bacterial wilt is the best way to control the disease. Early attempts to select resistant lines were made in Florida and North Carolina (Hume, 1903; Massey, 1903; Sherbakoff, 1919). Louisiana Pink was the first commercial variety with



partial resistance to bacterial wilt (Schmidt, 1938). In 1953 *Lycopersicon pimpinellifolium* (PI 127805A) was identified in Hawaii and since then, attempts have been made to incorporate resistance into other varieties and lines (Russell, 1978).

Recently, the Asian Vegetable Research and Development Center (AVRDC) in Taiwan has become one of the most important centers working in the field of tomato breeding for resistance to bacterial wilt (AVRDC, 1993). Tomato seeds in this study were provided by the center.

Considering breeding methods for disease resistance in tomato, the backcross method provides a precise way for this breeding objective (Allard, 1960). The method was first used in 1922 to develop a bunt resistant varieties in wheat. Breeding for disease resistance is an important complementary objective. It is of no value if a variety is resistant to certain disease and of poor yielding ability and lower quality. In this study, the genetics of bacterial wilt resistance as well as yield and some quality characters are considered. Thus, the objectives of this study were :

1. To study the heritability of bacterial wilt resistance and some yield and quality characters.
2. To study the degree of heterosis of F1 hybrid above the mid parental and better parental values.
3. To elucidate the correlation between characters and regression of yield components on yield.
4. To study the nature of resistance and to investigate the gene action and environmental influence on some agronomic characters.

Based on the findings obtained in these studies, some selection criteria can be established for future breeding programmes

CHAPTER II

LITERATURE REVIEW

Bacterial Wilt of Tomato

Bacterial wilt of tomato is a wide spread disease caused by *Burkholderia solanacearum* (Synonym *Pseudomonas solanacearum*) It is a major disease in areas where the pathogen is well established in the soil (Singh, 1987) The disease has a wide range of host plants including those that exhibit symptoms of the disease and others which are symptomless carriers

Result of investigations indicated that the neem trees *Azadirachta indica* in Australia has been affected by wilt disease and the causal organism was *P solanacearum* biovar 3 was isolated from the trees That was the first time neem to be regarded as bacterial wilt susceptible plants (Diatloff et al , 1993)

Although the disease is well known as a soil borne disease which is usually found in cultivated (Kelman, 1953) and newly cleared land (Graham et al , 1977), Machmud and Middleton (1991) showed that the pathogen could be a seed borne in the ground nut seeds They isolated *P solanacearum* from groundnut stem, podshell, seed coat and embryo The pathogen was found pathogenic to groundnut and tomato

Abdullah (1992) conducted a survey in Malaysia to determine the hosts of *P. solanacearum* and found that it was isolated from 25 diseased crop plants, weeds and ornamentals. Greenhouse inoculation studies showed all the isolates were pathogenic to tomato.

As far as invasion or penetration of the bacteria into the plant tissues is concerned, Kelman (1953) explained that the pathogen penetrates through wounds found in the host roots. These wounds usually occur when cultivating, transplanting or through mechanical injuries of insects and nematodes. He added that bacteria could penetrate uninjured roots if the soil moisture was high. He suggested that penetration of healthy roots occurred at the points of emergence of secondary and tertiary roots.

Existence of *P. solanacearum* in the host plants can be indicated by isolating the pathogen from plant tissues, mainly the vascular system. However, Prior et al. (1990) used Enzyme Linked Immuno-Sorbant Assay (ELISA) to investigate the spread of the bacterium through vascular system of the hosts.

The most characteristic symptoms of bacterial wilt disease caused by *P. solanacearum* in tomato is the dropping of lower leaves of the host followed by rapid wilting especially when the plants are young and succulent. Other symptoms includes, plant stunting and down curling of leaflets and petioles. Usually infected plants collapse quickly, or their vascular system becomes black in color. Large number of the bacterial cells are released into the surrounding soil from infected roots and thus invading subsequent crops (Kelman, 1953; Dixon, 1981).

Gilbert et al (1966) indicated that, severe infestation with spider mites *Tetranychus spp* may cause wilt symptoms similar to bacterial wilt Grimault and Prior (1993) proposed the extent of bacterial colonization as a criterion to quantify tolerance, complementary to absence of external wilt symptoms used in breeding programs for resistance

Breeding for Bacterial Wilt Resistance

Different degrees of susceptibilities of tomatoes to bacterial wilt can be recognized after the causative agent was determined (Earle, 1900) At the beginning of the 20th century, many attempts had been made to select for resistant varieties in USA, but there was no promising results (Hume, 1903, Massey, 1903, Sherbakoff, 1919) Louisiana Pink was the only commercial variety with partial resistance to bacterial wilt produced at North Carolina In field trials, Louisiana Pink and *Lycopersicon esculentum* T 414 (accession no PI 3814) from Puerto Rica showed high degree of resistance Crosses made between these accessions were considered as promising source of resistance to bacterial wilt (Schaub and Barer, 1944)

In Australia, Aberdeen (1946) tested a number of tomato varieties for resistance to bacterial wilt He found that strains derived from Louisiana Pink and T 414 were resistant Resistant lines from North Carolina were also found to be resistant in Hawaii However, the fruits of these lines were too small to make them of considerable commercial values

In Sri Lanka, Abeygunawardena and Siriwardena (1964) tested 49 tomato varieties and hybrids for resistance to bacterial wilt Among them, were improved lines from North Carolina They reported that four of the North Carolina lines together with the varieties Masterglobe and Rahangala,



were found to be the most resistant to bacterial wilt. The resistant varieties yielded more than the local susceptible ones. Among these genotypes a line which had shown resistance to *P. solanacearum* in the Philippines, but was found susceptible to the pathogen in Sri Lanka. Different physiological races of *P. solanacearum* were then suggested to be found in these countries.

Acosta et al. (1964) found that Louisiana Pink and *Lycopersicon pimpinellifolium* accession PI 127805a were resistant to bacterial wilt. In 1972, Varieties Venus and Saturn, derived from crosses between Louisiana Pink and PI 3814 and *L. esculentum* var. *cerasitorme* (PI 129080), respectively, were released in North Carolina (Henderson and Jenkins, 1972)). These two varieties were resistant in French Antilles (Daly, 1973) and Taiwan (Lin et al., 1974). Unfortunately, in India, the North Carolina lines were found to be unpromising and only one line namely CRA 66-A was resistant to bacterial wilt but the fruit size of this line was very small. (Rao et al., 1975)

Mew and Ho (1976) found that certain resistant tomatoes showed an unexpected degree of susceptibility to bacterial wilt when they were exposed to high inoculum densities.

In 1973 the Asian Vegetables Research and Development Center (AVRDC) initiated research on bacterial wilt of tomato. One of the main objectives of AVRDC was to develop resistant cultivars adaptable to tropical environments. In 1986, a highly reliable field screening method was devised and it has been accepted as the standard protocol for conducting this type of research. The most resistant germplasm in the AVRDC collection was L 285, a primitive type. North Carolina and Hawaii germplasms were also used in the breeding programs (Opena et al., 1990).



In Malaysia, Ho (1988) found that, in a field trial, the cultivar MT1 was highly resistant to *Pseudomonas solanacearum* while MT2, MT3, MT5, MT7, MT8, MT10 and MT11 were moderately resistant. Banting and MT9 were susceptible. The disease reached its peak nine weeks after transplanting. However, in 1990 and 1991 MARDI (Malaysian Agricultural Research and Development Center) noticed that varieties BL 355 and CLN 65 were highly resistant to bacterial wilt (AVRDC, 1991).

Genetic Control of Bacterial Wilt Resistance

The genetics of the bacterial wilt resistance in tomato was studied by many workers. Acosta et al. (1964) reported that resistance in Venus, Saturn and North Carolina lines are polygenically inherited. Acosta (1972) identified the resistant genes in *Lycopersicon pimpinellifolium* as partially dominant genes at the seedling stage. However, Singh, (1961) reported that these factors were recessive and polygenically inherited. Ferrer (1974) studied some lines derived from Louisiana Pink (PI) and concluded that inheritance of resistance was determined by a single gene or few genes. Graham and Yap (1976) in a diallel-cross study, reported that inheritance of wilt resistance in the variety VC-4, a Philippine's variety, was mainly due to additive gene action. Anais (1986) suggested that in line CRA 66, resistance was controlled by four to five dominant genes. A study by Sreelathaakumary et al. (1987) showed that the crosses between Louisiana Pink and its derivative lines, from North Caroline type resistance (*L. esculentum*) and *L. pimpinellifolium* (PI 127805 A) type resistance, revealed that resistance to bacterial wilt was due to a recessive type of gene action. They concluded that a complementary gene action behavior involving two separate gene systems was governing the nature of resistance. However, in general, the